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DATE: Wednesday, November 30, 2005

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		<i>DB=PGPB,USPT,USOC,EPAB,JPAB,DWPI,TDBD; PLUR=YES; OP=OR</i>	
<input type="checkbox"/>	L1	singh.in. and anthra\$	170
<input type="checkbox"/>	L2	L1 and iota\$	3
<input type="checkbox"/>	L3	yogendra	1740
<input type="checkbox"/>	L4	L3 and anthra\$	156
<input type="checkbox"/>	L5	L4 and iota\$	3
<input type="checkbox"/>	L6	iota\$.clm. and (anthrax\$ or anthra\$).clm.	12
<input type="checkbox"/>	L7	(khanna or hemant)in. and anthra\$	3682
<input type="checkbox"/>	L8	L7 and iota\$	7
<input type="checkbox"/>	L9	5,935,990.pn.	2

END OF SEARCH HISTORY

DOCUMENT-IDENTIFIER: US 20020048590 A1

TITLE: Targeting antigens to the MHC class I processing pathway with an anthrax toxin fusion protein

CLAIMS:

1. A vaccine capable of inducing an immune response in a mammal to a specific antigen wherein the vaccine comprises a unit dose of an anthrax protective antigen and said specific antigen bound to an anthrax protective antigen binding protein.
6. The vaccine of claim 1 wherein the anthrax protective antigen binding protein is the lethal factor of *Bacillus anthracis*.
7. The vaccine of claim 1 wherein the anthrax protective antigen binding protein comprises at least about the first 250 amino acid residues of the lethal factor of *Bacillus anthracis* and less than all of the amino acid residues of the lethal factor.
8. The vaccine of claim 1 wherein the molar ratio of protective antigen to the antigen bound to an anthrax protective antigen binding protein is greater than one.
9. A method of immunizing a mammal against an antigen which comprises administering a safe and effective amount of a vaccine comprising an anthrax protective antigen and said antigen bound to an anthrax protective antigen binding protein.
14. The method of claim 9 wherein the anthrax protective antigen binding protein is the lethal factor of *Bacillus anthracis*.
15. The method of claim 9 wherein the anthrax protective antigen binding protein comprises at least about the first 250 amino acid residues of the lethal factor of *Bacillus anthracis* and less than all of the amino acid residues of the lethal factor.
16. The method of claim 9 wherein the molar ratio of protective antigen to the antigen bound to an anthrax protective antigen binding protein is greater than one.
19. The method of claim 9 wherein the vaccine is administered in a unit dose that is between 10 to 500 nanograms of antigen bound to an anthrax protective antigen binding protein per kilogram of said mammal.
20. A method of inducing antigen presenting mammalian cells to present specific antigens on their cell membranes via the MHC class I processing pathway, comprising: i) selecting cells that can process and present specific antigens on their cell membranes via the MHC class I processing pathway; ii) contacting the cells with an anthrax protective antigen and said specific antigen bound to an anthrax protective antigen binding protein; and, iii) permitting the cells to internalize, process and present said specific antigen bound to an anthrax protective antigen binding protein on its cell membrane, forming a specific antigen presenting cell.
23. The method of claim 20 wherein the anthrax protective antigen binding protein is the lethal factor of *Bacillus anthracis*.
24. The method of claim 20 wherein the anthrax protective antigen binding protein comprises at least about the first 250 amino acid residues of the lethal factor of *Bacillus anthracis* and less than all of the

amino acid residues of the lethal factor.

25. The method of claim 20 wherein the molar ratio of protective antigen to the antigen bound to an anthrax protective antigen binding protein is greater than one.

27. A vaccine for inducing an immune response in a mammal to a specific antigen wherein the vaccine comprises a unit dose of a binary toxin protective antigen and the antigen bound to a binary toxin protective antigen binding protein wherein the binary toxin is selected from the group comprising iota toxin and anthrax toxin.

28. The vaccine of claim 27, wherein the binary toxin is iota toxin.

20030198651. 27 May 03. 23 Oct 03. Targeting antigens to the MHC class I processing pathway with an anthrax toxin fusion protein. Klimpel, Kurt, et al. 424/246.1; A61K039/07.

☐ 8. 20030198595. 17 May 02. 23 Oct 03. Use of bi-specific antibodies for pre-targeting diagnosis and therapy. Goldenberg, David M., et al. 424/1.49; 530/391.1 534/11 A61K051/00 C07K016/46.

☐ 9. 20030148409. 15 Oct 02. 07 Aug 03. Direct targeting binding proteins. Rossi, Edmund, et al. 435/7.23; 424/1.49 530/388.8 A61K051/00 G01N033/574 C07K016/30.

☐ 10. 20020048590. 09 May 01. 25 Apr 02. Targeting antigens to the MHC class I processing pathway with an anthrax toxin fusion protein. Klimpel, Kurt, et al. 424/246.1; A61K039/07.

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L5: Entry 1 of 4

File: PGPB

Apr 25, 2002

DOCUMENT-IDENTIFIER: US 20020048590-A1

TITLE: Targeting antigens to the MHC class I processing pathway with an anthrax toxin fusion protein

CLAIMS:

27. A vaccine for inducing an immune response in a mammal to a specific antigen wherein the vaccine comprises a unit dose of a binary toxin protective antigen and the antigen bound to a binary toxin protective antigen binding protein wherein the binary toxin is selected from the group comprising iota toxin and anthrax toxin.

28. The vaccine of claim 27, wherein the binary toxin is iota toxin.

*Iota-b
873aa*

IDS

Sellman 2001

Surand 1997

PA-Q987

*Iota-b
Berelle
et al 1993*

WEST

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L5: Entry 2 of 4

File: USPT

Apr 10, 2001

DOCUMENT-IDENTIFIER: US 6214602 B1

TITLE: Host cells for expression of clostridial toxins and proteins

CLAIMS:

3. The host cell of claim 2, wherein said clostridial proteins are selected from the group consisting of light chains of botulinal neurotoxins, heavy chains of botulinal neurotoxins, botulinal C3 protein, clostridial iota toxin Ia protein, and light and heavy chains of tetanus toxin.

AUTHORS' CORRECTIONS

Characterization of *Clostridium perfringens* Iota-Toxin Genes and Expression in *Escherichia coli*

SYLVIE PERELLE, MARYSE GIBERT, PATRICE BOQUET, AND MICHEL R. POPOFF

Laboratoire des Toxines Microbiennes, Institut Pasteur, 28 Rue du Dr. Roux, 75724 Paris Cedex 15, France

Volume 61, no. 12, p. 5147-5156: A DNA sequence mistake has been found at the end of the Ia gene. The five C-terminal amino acids of Ia are incorrect, and the Ia sequence is 67 amino acids longer. The Ia and Ib genes are separated by 40 noncoding nucleotides; the Ib sequence is not changed. The Ia sequence has been corrected in the EMBL data library and should appear as shown below:

```
1 MKKVNSISV FLILYLILTS SFPSYTYAQD LQIASNYITD RAFIERPEDF
51 LKDKENAIQW EKKEAERVEK NLDTLEKEAL ELYKKDSEQI SNYSQTRQYF
101 YDYQIESNPR EKEYKNLRNA ISKNKIDKPI NVYYFESPEK FAFNKEIRTE
151 NQNEISLEKF NELKETIQDK LFKQDGFQDV SLYEPGNGDE KPTPLLIHLK
201 LPKNTGMLPY INSNDVKTLI EQDYSIKIDK IVRIVIEGKQ YIKAEASIVN
251 SLDFKDDVSK GDLWGKENYS DWSNKLTPNE LADVNDYMRG GYTAINNYLI
301 SNGPLNNPNP ELDSKVNNIE NALKLTPIPS NLIVYRRSGP QEFGLTLTSP
351 EYDFNKIENI DAFKEKWEGK VITYPNFIST SIGSVNMSAF AKRKIILRLN
401 IPKDS PGAYL SAIPGYAGEY EVLLNHGSKF KINKVDSYKD GTVTKLILDA
451 TLIN*
```

Characterization of the Structural Elements in Lipid A Required for Binding of a Recombinant Fragment of Bactericidal/Permeability-Increasing Protein rBPI₂₃

HÉLÈNE GAZZANO-SANTORO, JAMES B. PARENT, PAUL J. CONLON, HERBERT G. KASLER,
CHAO-MING TSAI, DEBORAH A. LILL-ELGHANIAN, AND RAWLE I. HOLLINGSWORTH

Sepsis Research Department, XOMA Corporation, Berkeley, California 94710; Neurocrine Biosciences, San Diego, California 92121; Department of Health and Human Services, Center for Biologics Evaluation and Research, Food and Drug Administration, Bethesda, Maryland 20892; and Departments of Biochemistry and Chemistry, Michigan State University, East Lansing, Michigan 48824

Volume 63, no. 6, p. 2201-2205: We failed to cite the study by Holst et al. in which the original structure of *Escherichia coli* J5 lipid A was first reported (O. Holst, S. Müller-Loennies, B. Lindner, and H. Brade, *Eur. J. Biochem.* **214**:695-701, 1993).

- ☐ tr Q9KH41 _CLODI CdtB [cdtB] [Clostridium difficile]
☐ tr O32739 _CLODI ADP-ribosyltransferase [cdtB] [Clostridium diffi

Graphical overview of the alignments

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([?](#) Help) (use ScanProsite for more details about PROSITE matches)

Profile hits

Pfam hits

Matches on query sequence

Submission

Q46221
O86498
Q9KH41
O32739

Submission

Identity 0 25 50 75 100%

Alignments

tr Q46221 Iota toxin component Ib precursor [Clostridium
Q46221_CLOPE perfringens]

875
AA
align

Score = 73.2 bits (165), Expect = 7e-13
Identities = 23/23 (100%), Positives = 23/23 (100%)

Query: 1 DANTVGVSISAGYQNGFTGNITT 23
DANTVGVSISAGYQNGFTGNITT
Sbjct: 332 DANTVGVSISAGYQNGFTGNITT 354

tr O06498 Sb component [sbs] [Clostridium spiroforme] 879 AA
O06498_9MOLU

align

Score = 52.8 bits (117), Expect = 9e-07
Identities = 17/23 (73%), Positives = 17/23 (73%)

Query: 1 DANTVGVSISAGYQNGFTGNITT 23

General Search

DANT GV I YQNGFTG ITT
Sbjct: 336 DANTAGVAINIAYQNGFTGSITT 358

tr Q9KH41 CdtB [cdtB] [Clostridium difficile] 876 AA
Q9KH41_CLODI align

Score = 49.8 bits (110), Expect = 7e-06
Identities = 15/23 (65%), Positives = 18/23 (78%)

Query: 1 DANTVGVSISAGYQNGFTGNITT 23
+ NT GVS+ GYQNGFT N+TT
Sbjct: 333 ESNTAGVSVNVGYQNGFTANVTT 355

tr 032739 ADP-ribosyltransferase [cdtB] [Clostridium difficile] 876 AA
032739_CLODI align

Score = 49.8 bits (110), Expect = 7e-06
Identities = 15/23 (65%), Positives = 18/23 (78%)

Query: 1 DANTVGVSISAGYQNGFTGNITT 23
+ NT GVS+ GYQNGFT N+TT
Sbjct: 333 ESNTAGVSVNVGYQNGFTANVTT 355

Database: EXPASY/UniProtKB

Posted date: Nov 21, 2005 2:19 PM

Number of letters in database: 854,910,163

Number of sequences in database: 2,618,771

Lambda	K	H
0.349	0.280	1.64

Gapped

Lambda	K	H
0.294	0.110	0.610

Matrix: PAM30

Gap Penalties: Existence: 9, Extension: 1

Number of HSP's successfully gapped in prelim test: 0

length of query: 23

length of database: 854,910,163

effective HSP length: 14
effective length of query: 9
effective length of database: 818,247,369
effective search space: 7364226321
effective search space used: 7364226321
T: 16
A: 40
X1: 14 (7.0 bits)
X2: 35 (14.8 bits)
X3: 58 (24.6 bits)
S1: 40 (22.0 bits)
S2: 62 (29.5 bits)

Wallclock time: 2 seconds



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[Swiss-Prot](#)

CLUSTAL W (1.82) multiple sequence alignment

```

sp|P13423|PAG_BACAN      MKKRKVLIPLMALSTILVSSTGNLEVIQAEVKQENRLLNESESSSQGLLG
tr|Q68GS1|Q68GS1_BACAN  -----MEVKQENRLLNESESSSQGLLG
tr|Q52NH4|Q52NH4_BACAN  MKKRKVLIPLMALSTILVSSTGNLEVIQAEVKQENRLLNESESSSQGLLG
tr|Q4ZE94|Q4ZE94_BACAN  -----

sp|P13423|PAG_BACAN      YYFSDLNFQAPMVVTSSTTGDLSPSSELENIPSENQYFQSAIWSGFIKV
tr|Q68GS1|Q68GS1_BACAN  YYFSDLNFQAPMVVTSSTTGDLSPSSELENIPSENQYFQSAIWSGFIKV
tr|Q52NH4|Q52NH4_BACAN  YYFSDLNFQAPMVVTSSTTGDLSPSSELENIPSENQYFQSAIWSGFIKV
tr|Q4ZE94|Q4ZE94_BACAN  -----

sp|P13423|PAG_BACAN      KKSDEYTFATSADNHVTMWVDDQEVINKASNSNKIRLEKGRLYQIKIQYQ
tr|Q68GS1|Q68GS1_BACAN  KKSDEYTFATSADNHVTMWVDDQEVINKASNSNKIRLEKGRLYQIKIQYQ
tr|Q52NH4|Q52NH4_BACAN  KKSDEYTFATSADNHVTMWVDDQEVINKASNSNKIRLEKGRLYQIKIQYQ
tr|Q4ZE94|Q4ZE94_BACAN  -----

sp|P13423|PAG_BACAN      RENPTEKGLDFKLYWTDSONKKEVISSDNLQLPELKQKSSNSRKKRSTSA
tr|Q68GS1|Q68GS1_BACAN  RENPTEKGLDFKLYWTDSONKKEVISSDNLQLPELKQKSSNSRKKRSTSA
tr|Q52NH4|Q52NH4_BACAN  RENPTEKGLDFKLYWTDSONKKEVISSDNLQLPELKQKSSNSRKKRSTSA
tr|Q4ZE94|Q4ZE94_BACAN  -----

sp|P13423|PAG_BACAN      GPTVPDRDNDGIPDSLEVEGYTVDVKNKRTFLSPWISNIHEKKGLTKYKS
tr|Q68GS1|Q68GS1_BACAN  GPTVPDRDNDGIPDSLEVEGYTVDVKNKRTFLSPWISNIHEKKGLTKYKS
tr|Q52NH4|Q52NH4_BACAN  GPTVPDRDNDGIPDSLEVEGYTVDVKNKRTFLSPWISNIHEKKGLTKYKS
tr|Q4ZE94|Q4ZE94_BACAN  ---VPDRDNDGIPDSLEVEGYTVDVKNKRTFLSPWISNIHEKKGLTKYKS
                        *****

sp|P13423|PAG_BACAN      SPEKWSTASDPYSDFEKV TGRIDKNVSPEARHPLVAAYPIVHVDMENIIL
tr|Q68GS1|Q68GS1_BACAN  SPEKWSTASDPYSDFEKV TGRIDKNVSPEARHPLVAAYPIVHVDMENIIL
tr|Q52NH4|Q52NH4_BACAN  SPEKWSTASDPYSDFEKV TGRIDKNVSPEARHPLVAAYPIVHVDMENIIL
tr|Q4ZE94|Q4ZE94_BACAN  SPEKWSTASDPYSDFEKV TGRIDKNVSPEARHPLVAAYPIVHVDMENIIL
                        *****

sp|P13423|PAG_BACAN      SKNEDQSTQNTDSQTRTISKNTSTSRHTHTSEVHGNAEVHASFFDIGGSVS
tr|Q68GS1|Q68GS1_BACAN  SKNEDQSTQNTDSQTRTISKNTSTSRHTHTSEVHGNAEVHASFFDIGGSVS
tr|Q52NH4|Q52NH4_BACAN  SKNEDQSTQNTDSQTRTISKNTSTSRHTHTSEVHGNAEVHASFFDIGGSVS
tr|Q4ZE94|Q4ZE94_BACAN  SKNEDQSTQNTDSQTRTISKNTSTSRHTHTSEVHGNAEVHASFFDIGGSVS
                        *****

sp|P13423|PAG_BACAN      AGFSNSNSSTVAIDHSLSLAGERTWAETMGLNTADTARLNANIRYVNTGT
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tr|Q52NH4|Q52NH4_BACAN  AGFSNSNSSTVAIDHSLSLAGERTWAETMGLNTADTARLNANIRYVNTGT
tr|Q4ZE94|Q4ZE94_BACAN  AGFSNSNSSTVAIDHSLSLAGERTWAETMGLNTADTARLNANIRYVNTGT
                        *****

sp|P13423|PAG_BACAN      APIYNVLPPTTSLVLGKNQTLATIKAKENQLSQILAPNNYYP SKNLAPIAL
tr|Q68GS1|Q68GS1_BACAN  APIYNVLPPTTSLVLGKNQTLATIKAKENQLSQILAPNNYYP SKNLAPIAL
tr|Q52NH4|Q52NH4_BACAN  APIYNVLPPTTSLVLGKNQTLATIKAKENQLSQILAPNNYYP SKNLAPIAL
tr|Q4ZE94|Q4ZE94_BACAN  APIYNVLPPTTSLVLGKNQTLATIKAKENQLSQILAPNNYYP SKNLAPIAL
                        *****

sp|P13423|PAG_BACAN      NAQDDFSSTPITMNYNQFLELEKTKQLRLD TDQVYGN IATYNFENGRVRV
tr|Q68GS1|Q68GS1_BACAN  NAQDDFSSTPITMNYNQFLELEKTKQLRLD TDQVYGN IATYNFENGRVRV
tr|Q52NH4|Q52NH4_BACAN  NAQDDFSSTPITMNYNQFLELEKTKQLRLD TDQVYGN IATYNFENGRVRV

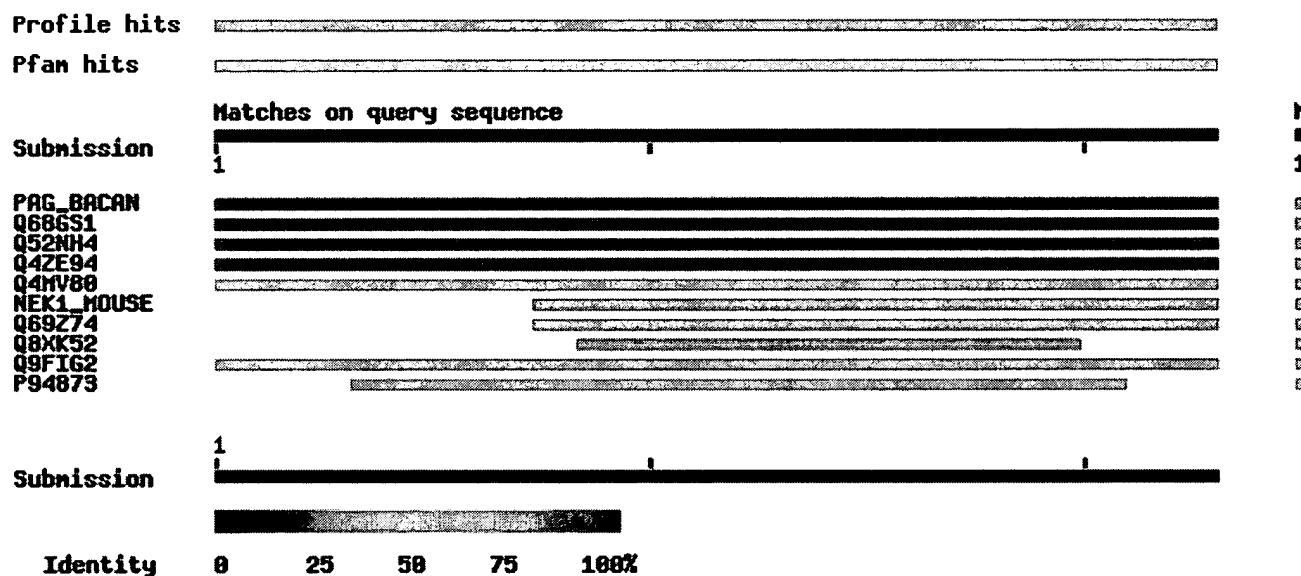
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tr	Q4ZE94	Q4ZE94_BACAN	NAQDDFSSTPITMNYNQFLELEKTKQLRLD TDQVYGN IATYNFENGRVRV *****
sp	P13423	PAG_BACAN	DTGSNWSEVLPQIQETTARI IIFNGKDLNLVERRIAAVNPSPLETTKPDM
tr	Q68GS1	Q68GS1_BACAN	DTGSNWSEVLPQIQETTARI IIFNGKDLNLVERRIAAVNPSPLETTKPDM
tr	Q52NH4	Q52NH4_BACAN	DTGSNWSEVLPQIQETTARI IIFNGKDLNLVERRIAAVNPSPLETTKPDM
tr	Q4ZE94	Q4ZE94_BACAN	DTGSNWSEVLPQIQETTARI IIFNGKDLNLVERRIAAVNPSPLETTKPDM *****
sp	P13423	PAG_BACAN	TLKEALKIAFGFNEPNGNLQYQGKDITEFDNF DQQT SQNIKNQLAELNA
tr	Q68GS1	Q68GS1_BACAN	TLKEALKIAFGFNEPNGNLQYQGKDITEFDNF DQQT SQNIKNQLAELNA
tr	Q52NH4	Q52NH4_BACAN	TLKEALKIAFGFNEPNGNLQYQGKDITEFDNF DQQT SQNIKNQLAELNA
tr	Q4ZE94	Q4ZE94_BACAN	TLKEALKIAFGFNEPNGNLQYQGKDITEFDNF DQQT SQNIKNQLAELNA *****
sp	P13423	PAG_BACAN	TNIYTVLDKIKLNAKMNILIRDKRFHYDRNNI AVGADES VVKEAHREVIN
tr	Q68GS1	Q68GS1_BACAN	TNIYTVLDKIKLNAKMNILIRDKRFHYDRNNI AVGADES VVKEAHREVIN
tr	Q52NH4	Q52NH4_BACAN	TNIYTVLDKIKLNAKMNILIRDKRFHYDRNNI AVGADES VVKEAHREVIN
tr	Q4ZE94	Q4ZE94_BACAN	TNIYTVLDKIKLNAKMNILIRDKRFHYDRNNI AVGADES VVKEAHREVIN *****
sp	P13423	PAG_BACAN	SSTEGLLLLNIDKDIRKILSGYIVEIEDTEGLKEVINDRYDMLN ISSLRQD
tr	Q68GS1	Q68GS1_BACAN	SSTEGLLLLNIDKDIRKILSGYIVEIEDTEGLKEVINDRYDMLN ISSLRQD
tr	Q52NH4	Q52NH4_BACAN	SSTEGLLLLNIDKDIRKILSGYIVEIEDTEGLKEVINDRYDMLN ISSLRQD
tr	Q4ZE94	Q4ZE94_BACAN	SSTEGLLLLNIDKDIRKILSGYIVEIEDTEGLKEVINDRYDMLN ISSLRQD *****
sp	P13423	PAG_BACAN	GKTFIDFKKYNDKLPLYISNP NYKVNVYAVTKENTI INPSENGDTSTNGI
tr	Q68GS1	Q68GS1_BACAN	GKTFIDFKKYNDKLPLYISNP NYKVNVYAVTKENTI INPSENGDTSTNGI
tr	Q52NH4	Q52NH4_BACAN	GKTFIDFKKYNDKLPLYISNP NYKVNVYAVTKENTI INPSENGDTSTNGI
tr	Q4ZE94	Q4ZE94_BACAN	GKTFIDFKKYNDKLPLYISNP NYKVNVYAVTKENTI INPSENGDTSTNGI *****
sp	P13423	PAG_BACAN	KKILIFS KKG YEIG
tr	Q68GS1	Q68GS1_BACAN	KKILIFS KKG YEIG
tr	Q52NH4	Q52NH4_BACAN	KKILIFS KKG YEIG
tr	Q4ZE94	Q4ZE94_BACAN	KKILIFS KKG YEIG *****

- ☒ sp P13423 PAG_BACAN Protective antigen precursor (PA) (PA-83) (P.
- ☒ tr Q68GS1 _BACAN Protective antigen [Bacillus anthracis]
- ☒ tr Q52NH4 _BACAN Protective antigen [pag] [Bacillus anthracis]
- ☒ tr Q4ZE94 _BACAN Protective antigen (Fragment) [pa] [Bacillus ant
- ☐ tr Q4MV80 _BACCE Protective antigen [BCE_G9241_pBC218_0026] [Baci
- ☐ sp P51954 NEK1_MOUSE Serine/threonine-protein kinase Nek1 (EC 2..
- ☐ tr Q69Z74 _MOUSE MKIAA1901 protein (Fragment) [Nek1] [Mus musculu
- ☐ tr Q8XK52 _CLOPE Probable endo-1,4-beta-xylanase [CPE1551] [Clost
- ☐ tr Q9FIG2 _ARATH Serine protease-like protein (Hypothetical prote
- ☐ tr P94873 _LYSLA Alpha-aminoadipyl-cysteinyl-valine synthetase [p

Graphical overview of the alignments

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 (🔗 Help) (use ScanProsite for more details about PROSITE matches)



Alignments

sp P13423 Protective antigen precursor (PA) (PA-83) (PA83) (Anthrax 764
 PAG_BACAN toxins AA
 translocating protein) [Contains: Protective antigen align
 PA-20 (PA20); Protective antigen PA-63 (PA63)] [pagA]
 [Bacillus anthracis]

Score = 74.4 bits (168), Expect = 3e-13
 Identities = 23/23 (100%), Positives = 23/23 (100%)

Query: 1 THTSEVHGNAEVHASFFDIGGSV 23
THTSEVHGNAEVHASFFDIGGSV
Sbjct: 327 THTSEVHGNAEVHASFFDIGGSV 349

tr Q68GS1 Protective antigen [Bacillus anthracis] 736 AA
Q68GS1_BACAN align

Score = 74.4 bits (168), Expect = 3e-13
Identities = 23/23 (100%), Positives = 23/23 (100%)

Query: 1 THTSEVHGNAEVHASFFDIGGSV 23
THTSEVHGNAEVHASFFDIGGSV
Sbjct: 299 THTSEVHGNAEVHASFFDIGGSV 321

tr Q52NH4 Protective antigen [pag] [Bacillus anthracis] 764 AA
Q52NH4_BACAN align

Score = 74.4 bits (168), Expect = 3e-13
Identities = 23/23 (100%), Positives = 23/23 (100%)

Query: 1 THTSEVHGNAEVHASFFDIGGSV 23
THTSEVHGNAEVHASFFDIGGSV
Sbjct: 327 THTSEVHGNAEVHASFFDIGGSV 349

tr Q4ZE94 Protective antigen (Fragment) [pa] [Bacillus anthracis] 561 AA
Q4ZE94_BACAN align

Score = 74.4 bits (168), Expect = 3e-13
Identities = 23/23 (100%), Positives = 23/23 (100%)

Query: 1 THTSEVHGNAEVHASFFDIGGSV 23
THTSEVHGNAEVHASFFDIGGSV
Sbjct: 124 THTSEVHGNAEVHASFFDIGGSV 146

CLUSTAL W (1.81) multiple sequence alignment

```

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          MNIIQIKNVFSFLTTLTAMISQTLSYNVYAQTTTQNDTNQKEEITNENTLS

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          NGLMGYYFADEHFKDLELMAPIKNGDLKFEEKKVDKLLTEDNSSIKSIR

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          TGRIIPSEDGEYILSTDRNDVLMQINAKGDIKTLKVNMMKKGQAYNIRI

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          IQDKNLGSIDNLSVPKLYWELNGNKTVIPEENLFFRDYSKIDENDPFIP

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          NNFFDVRFFSAAWEDEDLDTDNDNI PDAYEKNGYTIKDSIAVKWNSFA

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          QGYKKYVSSYLESNTAGDPYTDYQKASGSIDKAIKLEARDPLVAAYPVV

unk|VIRT2310|Blast_submission -----DANTVGVSISAGYQNGFT
tr|Q46221|Q46221_CLOPE          VGMENLIISTNEHASSDQGKTVSRATTNSKTDANTVGVSISAGYQNGFT
                                *****

unk|VIRT2310|Blast_submission NITT-----
tr|Q46221|Q46221_CLOPE          NITTSYSHTTDNSTAVQDSNGESWNTGLSINKGESAYINANVRYNTGT
                                ****

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          PMYKVTPTTNLVLDGETLATIKAQDNQIGNNLSPNETYPPKGLSPLALN

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          MDQFNARLIPINYDQLKKLDSGKQIKLETTQVSGNYGTKNSQGQIITEG

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          SWSNYISQIDSVSASIIILDTGSQTFERRVAAKEQGNPEDKTPEITIGEA

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          KKAFSATKNGELLYFNGIPIDESCVELIFDDNTSEIIKEQLKYLDKKI

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          NVKLERGMNILIKVPSYFTNFDEYNNFPASWSNIDTKNQDGLQSVANKL

unk|VIRT2310|Blast_submission -----
tr|Q46221|Q46221_CLOPE          GETKIIIPMSKLPYKRYVFSGYSKDPSTSNSITVNIKSKEQKTDYLPV

unk|VIRT2310|Blast_submission -----

```

tr Q46221 Q46221_CLOPE	KDYTKFSYEFETTGDSSDIEITLTSSGVIFLDNLSITELNSTPEILKE
unk VIRT2310 Blast_submission	-----
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NCBI BLAST program reference [PMID:9254694]:
Altschul S.F., Madden T.L., Schäffer A.A., Zhang J., Zhang Z., Miller W., Lipman D.J. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. Nucleic Acids Res. 25:3389-3402(1997).

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Query: 23 AA

Date run: 2005-11-30 16:03:15 UTC+0100 on sib-gml.unil.ch

Program: NCBI BLASTP 1.5.4-Paracel [2003-06-05]

Database: EXPASY/UniProtKB

2,618,771 sequences; 854,910,163 total letters

UniProt Knowledgebase Release 6.5 consists of:

UniProtKB/Swiss-Prot Release 48.5 of 22-Nov-2005: 199607 entries

UniProtKB/TrEMBL Release 31.5 of 22-Nov-2005: 2406391 entries

List of potentially matching sequences

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Db	AC	Description
----	----	-------------

- | | | |
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| <input type="checkbox"/> | tr O06498 | _9MOLU Sb component [sbs] [Clostridium spiroforme] |



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SV      X73562.1
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DT      18-APR-2005 (Rel. 83, Last updated, Version 15)
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OC      Bacteria; Firmicutes; Clostridia; Clostridiales; Clostridiaceae;
OC      Clostridium.
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RA      Perelle S., Gibert M., Boquet P., Popoff M.R.;
RT      "Characterization of Clostridium perfringens iota toxin genes and
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RL      Infect. Immun. 61(12):5147-5156(1993).
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RN      [2]
RC      revised by [3]    MAT
RA      Popoff M.R.;
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RL      Submitted (30-APR-1993) to the EMBL/GenBank/DDBJ databases.
RL      M.R. Popoff, Institut Pasteur, 28 rue du Dr. Roux, Toxines Microbiennes,
RL      75015 Paris Cedex 15, FRANCE
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RN      [3]
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RT      ;
RL      Submitted (03-AUG-1995) to the EMBL/GenBank/DDBJ databases.
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entry Q46221

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[\[Keywords\]](#) [\[Features\]](#) [\[Sequence\]](#) [\[Tools\]](#)

Note: most headings are clickable, even if they don't appear as links. They link to the user manual or other documents.

Entry information

Entry name	Q46221_CLOPE
Primary accession number	Q46221
Secondary accession numbers	None
Entered in TrEMBL in	Release 01, November 1996
Sequence was last modified in	Release 01, November 1996
Annotations were last modified in	Release 24, June 2003
Name and origin of the protein	
Protein name	Iota toxin component Ib [Precursor]
Synonyms	None
Gene name	None
From	Clostridium perfringens [TaxID: 1502]
Taxonomy	Bacteria; Firmicutes; Clostridia; Clostridiales; Clostridiace Clostridium.

References

- [1] NUCLEOTIDE SEQUENCE.
STRAIN=NCIB 10748;
PubMed=8225592 [NCBI, ExPASy, EBI, Israel, Japan]
Perelle S., Gibert M., Boquet P., Popoff M.R.;
"Characterization of Clostridium perfringens iota toxin genes and expression in Escherichia coli.";
Infect. Immun. 61:5147-5156(1993).
- [2] NUCLEOTIDE SEQUENCE.
STRAIN=NCIB 10748;
Popoff M.R.;
Submitted (AUG-1995) to the EMBL/GenBank/DDBJ databases.

Comments

None

Cross-references

EMBL	X73562; CAA51960.1; -; Genomic_DNA.	[EMBL / GenBank / DDBJ] [CodingSequence]
PIR	I40862; I40862.	
HSSP	P13423; 1ACC. [HSSP ENTRY / PDB]	

GO GO:0005576; Cellular component: extracellular region (*inferred from electronic annotation*).

GO GO:0009405; Biological process: pathogenesis (*inferred from electronic annotation*).

QuickGo view.

InterPro IPR003896; Anthrax_toxinB.
IPR011658; PA14.
Graphical view of domain structure.

Pfam PF03495; Binary_toxB; 1.
PF07691; PA14; 1.
Pfam graphical view of domain structure.

PRINTS PR01391; BINARYTOXINB.

ProDom [Domain structure / List of seq. sharing at least 1 domain]

HOGENOM [Family / Alignment / Tree]

ProtoMap Q46221.

PRESAGE Q46221.

ModBase Q46221.

SWISS-2DPAGE Get region on 2D PAGE.

UniRef View cluster of proteins with at least 50% / 90% / 100% identity.

Keywords**Signal.****Features**

Feature table viewer

Key	From	To	Length	Description
SIGNAL	34	39	6	Potential.
CHAIN	212	875	664	iota toxin component Ib.

Sequence information

Length: **875 AA** [This is the length of the unprocessed precursor]

Molecular weight: **98469 Da** [This is the MW of the unprocessed precursor]

CRC64: **C9AE092CD3818921** is a checksum on the sequence

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Q
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PeptideCutter, Dotlet (Java)



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Nucleotide

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Range: from to ☐ Reverse complemented strand Features: ☐ SNP ☐ CDD ☒

☐ 1: X73562. Reports *C.perfringens* DNA...[gi:929031]

[Links](#)

LOCUS CPITOXIAB 5745 bp DNA linear BCT 18-APR-2005

DEFINITION *C.perfringens* DNA for iota toxin polypeptides Ia and Ib.

ACCESSION X73562

VERSION X73562.1 GI:929031

KEYWORDS iota toxin; iota toxin Ia; iota toxin Ib.

SOURCE *Clostridium perfringens*

ORGANISM *Clostridium perfringens*
Bacteria; Firmicutes; Clostridia; Clostridiales; Clostridiaceae;
Clostridium.

REFERENCE 1

AUTHORS Perelle,S., Gibert,M., Boquet,P. and Popoff,M.R.

TITLE Characterization of *Clostridium perfringens* iota-toxin genes and expression in *Escherichia coli*

JOURNAL Infect. Immun. 61 (12), 5147-5156 (1993)

PUBMED [8225592](#)

REFERENCE 2

AUTHORS Popoff,M.R.

TITLE Direct Submission

JOURNAL Submitted (30-APR-1993) M.R. Popoff, Institut Pasteur, 28 rue du Dr. Roux, Toxines Microbiennes, 75015 Paris Cedex 15, FRANCE

REMARK revised by [3] MAT

REFERENCE 3 (bases 1 to 5745)

AUTHORS Popoff,M.R.

TITLE Direct Submission

JOURNAL Submitted (03-AUG-1995) M.R. Popoff, Institut Pasteur, 28 rue du Dr. Roux, Toxines Microbiennes, 75015 Paris Cedex 15, FRANCE

COMMENT On Aug 5, 1995 this sequence version replaced [gi:414653](#).

FEATURES

Location/Qualifiers

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Go to: [General](#) [Description](#) [References](#) [Additional](#) [Sequence](#)

General Information

Primary Accession # X73562
 Accession # X73562
 Entry Name EMBL:CPITOXIAB
 Molecule Type genomic DNA
 Sequence Length 5745
 Entry Division PRO
 Sequence Version X73562.1
 Creation Date 04-NOV-1993
 Modification Date 18-APR-2005

Description

Description C.perfringens DNA for iota toxin polypeptides Ia and Ib
 Keywords iota toxin; iota toxin Ia; iota toxin Ib.;
 Organism Clostridium perfringens
 Organism Classification Bacteria; Firmicutes; Clostridia; Clostridiales; Clostridiaceae; Clostridium.

References

1. Perelle,S.; Gibert,M.; Boquet,P.; Popoff,M.R.;
Characterization of Clostridium perfringens iota toxin gene expression in Escherichia coli
 Infect. Immun. 61(12):5147-5156 (1993)
 Pubmed [8225592](#)
2. Popoff,M.R.; Submitted (30-APR-1993) to the EMBL/GenBank/DDBJ databases.
 Popoff, Institut Pasteur, 28 rue du Dr. Roux, Toxines Microbiennes, 75015 Paris C
 FRANCE
3. Popoff,M.R.; Submitted (03-AUG-1995) to the EMBL/GenBank/DDBJ databases
 Popoff, Institut Pasteur, 28 rue du Dr. Roux, Toxines Microbiennes, 75015 Paris C
 FRANCE
 Position 1-5745

Additional Information

Features

Key	Location	Qualifier	Value
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		mol_type	genomic DNA
		organism	Clostridium perfringens

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Sequence

Characteristics

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L2: Entry 2 of 25

File: USPT

Jul 15, 2003

DOCUMENT-IDENTIFIER: US 6592872 B1

TITLE: Targeting antigens to the MHC class I processing pathway with an anthrax toxin fusion protein

Brief Summary Text (97):

In general, for cloning and expression of PA, the same methods as described for antigen-APABP can be used by one skilled in the art. Genes that encode wild type or mutated proteins can be cloned and expressed by methods known to those skilled in the art, as described above. For example, the gene encoding protein Ib of the Clostridium perfringens iota toxin can be cloned and expressed for use in the present invention according the methods described herein, or by methods known to those skilled in the art. The present invention uses an isolated nucleic acid in expression vector pYS5 that encodes the PA protein, as described in Example 2.

Other Reference Publication (4):

Sirard, Jean-Claude, et al. (1997) "A Recombinant Bacillus anthracis Strain Producing the Clostridium perfringens Ib Component Induces Protection against Iota Toxins", Infection and Immunity, 65(6): 2029-2033.

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TITLE: Targeting antigens to the MHC class I processing pathway with an anthrax toxin fusion protein

Brief Summary Text (97):

In general, for cloning and expression of PA, the same methods as described for antigen-APABP can be used by one skilled in the art. Genes that encode wild type or mutated proteins can be cloned and expressed by methods known to those skilled in the art, as described above. For example, the gene encoding protein Ib of the Clostridium perfringens iota toxin can be cloned and expressed for use in the present invention according the methods described herein, or by methods known to those skilled in the art. The present invention uses an isolated nucleic acid in expression vector pYS5 that encodes the PA protein, as described in Example 2.

Other Reference Publication (4):

Sirard, Jean-Claude, et al. (1997) "A Recombinant Bacillus anthracis Strain Producing the Clostridium perfringens Ib Component Induces Protection against Iota Toxins", Infection and Immunity, 65(6): 2029-2033.

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OM protein - protein search, using sw model

Run on: July 30, 2003, 16:45:27 / Search time 95 Seconds
(without alignments)
62.476 Million cell updates/sec

Title: US-09-821-348-2
Perfect score: 117
Sequence: 1 VGVSIAGYQNGFTGNTTSAGP 23

Scoring table: BLOSUM62
Gapop 10.0, Gapext 0.5

Searched: 830525 seqs, 258052604 residues

Total number of hits satisfying chosen parameters: 830525

Minimum DB seq length: 0

Maximum DB seq length: 200000000

Post-processing: Minimum Match 0%

Maximum Match 100%

Listing first 45 summaries

Database :

1: sp_archea.*
2: sp_bacteria.*
3: sp_fungi.*
4: sp_human.*
5: sp_invertebrate.*
6: sp_mammal.*
7: sp_mhc.*
8: sp_organelle.*
9: sp_plant.*
10: sp_protist.*
11: sp_rodent.*
12: sp_virus.*
13: sp_vertebrate.*
14: sp_unclassified.*
15: sp_virus.*
16: sp_bacteriap.*
17: sp_archeap.*

Pred. No. is the number of results predicted by chance to have a score greater than or equal to the score of the result being printed, and is derived by analysis of the total score distribution.

SUMMARIES

Result No.	Score	Query Match %	Length	ID	Description
1	101	86.3	875	2 Q46221	Q46221 clostridium
2	79	67.5	876	2 Q32739	Q32739 clostridium
3	79	67.5	876	2 Q9841	Q9841 clostridium
4	72	61.5	879	2 Q06498	Q06498 clostridium
5	54	46.2	551	16 Q8A40	Q8A40 escherichia
6	54	46.2	551	16 Q8B25	Q8B25 escherichia
7	52	44.4	360	11 Q8K04	Q8K04 mus musculus
8	52	44.4	362	3 Q9HE29	Q9HE29 emezicella
9	52	44.4	538	16 Q8U730	Q8U730 agrobacteri
10	52	44.4	852	11 Q8BZG1	Q8BZG1 mus musculus
11	51	43.6	689	11 Q8BKG1	Q8BKG1 mus musculus
12	51	43.6	804	11 Q9NC2	Q9NC2 mus musculus
13	51	43.6	906	11 Q9FM5	Q9FM5 mus musculus
14	51	43.6	965	11 Q9PW2	Q9PW2 mus musculus
15	51	43.6	1160	11 Q9WU1	Q9WU1 mus musculus
16	51	43.6	1160	11 Q9R564	Q9R564 mus musculus

17	51	43.6	1198	11 Q99PM6	Q99PM6 mus musculus
18	51	43.6	1198	11 Q924G8	Q924G8 mus musculus
19	51	43.6	1987	11 Q99PB3	Q99PB3 mus musculus
20	51	43.6	3600	10 Q9SA64	Q9SA64 arabidopsis
21	50.5	43.2	434	16 Q983K6	Q983K6 rhizobium 1
22	50	42.7	129	16 Q8YQU6	Q8YQU6 anabaena sp
23	50	42.7	528	3 P87239	P87239 schizosacch
24	50	42.7	537	16 Q8F124	Q8F124 escherichia
25	50	42.7	1341	16 Q8UAU1	Q8UAU1 agrobacteri
26	49	41.9	357	16 Q8KU14	Q8KU14 vibrio chol
27	49	41.9	482	11 Q8BU52	Q8BU52 mus musculus
28	48.5	41.5	228	17 Q9FWM4	Q9FWM4 methanococ
29	48	41.0	114	2 Q9RHG4	Q9RHG4 microcystis
30	48	41.0	160	4 Q8NC35	Q8NC35 homo sapien
31	48	41.0	228	11 Q8COL2	Q8COL2 mus musculus
32	48	41.0	253	16 Q8ZQCS	Q8ZQCS salmonella
33	48	41.0	253	16 Q8Z804	Q8Z804 salmonella
34	48	41.0	315	2 Q9X588	Q9X588 neisseria f
35	48	41.0	381	11 Q8BX42	Q8BX42 mus musculus
36	48	41.0	537	5 Q9Y154	Q9Y154 drosophila
37	48	41.0	545	5 Q9VA35	Q9VA35 drosophila
38	48	41.0	653	4 Q96JN4	Q96JN4 homo sapien
39	48	41.0	663	2 Q8RR60	Q8RR60 rhizobium s
40	48	41.0	727	4 Q9UDR4	Q9UDR4 homo sapien
41	48	41.0	835	4 Q9ULP5	Q9ULP5 homo sapien
42	48	41.0	1323	2 Q87018	Q87018 helicobacte
43	48	41.0	1452	4 Q9H4A0	Q9H4A0 homo sapien
44	48	41.0	1512	4 Q9H4A1	Q9H4A1 homo sapien
45	47.5	40.6	3659	16 Q98LW6	Q98LW6 rhizobium 1

ALIGNMENTS

RESULT 1

Q46221 ID Q46221 PRELIMINARY; PRT; 875 AA.
AC Q46221, 1996 (TrEMBLrel. 01, Created)
DT 01-NOV-1996 (TrEMBLrel. 01, Last sequence update)
DT 01-JUN-2002 (TrEMBLrel. 21, Last annotation update)
DE Iota toxin component ib precursor.
OS Clostridium perfringens.
OC Bacteria; Firmicutes; Clostridia; Clostridiales; Clostridiaceae; Clostridium.
OK NCBI_TaxID=1502;
RN [1]
RP SEQUENCE FROM N.A.
RC STRAIN=NCIB 10748;
RX MEDLINE=94041637; PubMed=8225592;
RA Pirelle S., Gilbert M., Boquet P., Popoff M.R.;
RT "Characterisation of Clostridium perfringens Iota toxin genes and
RL expression in Escherichia coli.";
RN Infect. Immun. 61:5147-5156(1993).
RN [2]
RP SEQUENCE FROM N.A.
RC STRAIN=NCIB 10748;
RA Popoff M.R.;
RL Submitted (AUG-1995) to the EMBL/GenBank/DBJ databases.
DR EMBL; J13562; CAA31960.1; -.
DR HSP; F13423; IACC.
DR InterPro; IPR003896; Anthrax toxinB.
DR Pfam; PF03495; Binary toxinB; 1.
DR PRINTS; PR01391; BINARYTOXINB.
KW Signal.
FT CHAIN 34 38 POTENTIAL.
FT SIGNAL 212 875 IOTA TOXIN COMPONENT IB.
SQ SEQUENCE 875 AA; 98468 MW; C9AE02CD3818921 CRC64;

Query Match 86.3%; Score 101; DB 2; Length 875;
Best Local Similarity 100.0%; Pred. No. 1e-05;
Matches 20; Conservative 0; Mismatches 0; Indels 0; Gaps 0;



Infect. Immun., 12 1993, 5147-5156, Vol 61, No. 12
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Characterization of *Clostridium perfringens* iota-toxin genes and expression in *Escherichia coli* [published erratum appears in Infect Immun 1995 Dec;63(12):4967]

S Perelle, M Gibert, P Boquet and MR Popoff

Laboratoire des Toxines Microbiennes, Institut Pasteur, Paris, France.

The iota toxin which is produced by *Clostridium perfringens* type E, is a binary toxin consisting of two independent polypeptides: Ia, which is an ADP-ribosyltransferase, and Ib, which is involved in the binding and internalization of the toxin into the cell. Two degenerate oligonucleotide probes deduced from partial amino acid sequence of each component of *C. perfringens* type E plasmid DNA. Two genes, in the same orientation, coding for Ia (387 amino acids) and Ib (875 amino acids) and separated by 243 noncoding nucleotides were identified. A predicted signal peptide was found for each component, and the secreted Ib displays two domains, the propeptide (172 amino acids) and the mature protein (664 amino acids). The Ia gene has

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been expressed in *Escherichia coli* and *C. perfringens*, under the control of its own promoter. The recombinant polypeptide obtained was recognized by Ia antibodies and ADP-ribosylated actin. The expression of the Ib gene was obtained in *E. coli* harboring a recombinant plasmid encompassing the putative promoter upstream of the Ia gene and the Ia and Ib genes. Two residues which have been found to be involved in the NAD⁺ binding site of diphtheria and pseudomonas toxins are conserved in the predicted Ia sequence (Glu-14 and Trp-19). The predicted amino acid Ib sequence shows 33.9% identity with and 54.4% similarity to the protective antigen of the anthrax toxin complex. In particular, the central region of Ib, which contains a predicted transmembrane segment (Leu-292 to Ser-308), presents 45% identity with the corresponding protective antigen sequence which is involved in the translocation of the toxin across the cell membrane.

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